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Tubular handling of bicarbonate in dogs with experimental renal failure

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Tubular handling of bicarbonate in dogs with experimental renal failure. In this study, micropuncture experiments were performed to examine the segmental reabsorption of bicarbonate and chloride in the normal dog kidney (stage 1) and in the remnant kidney (stage 3) of dogs with experimental renal failure. The protocol consisted of three phases: *(1)* 3% extracellular fluid volume expansion, *(2)* infusion of Ringer's solution containing 90 mM of sodium bicarbonate, and *(3)* infusion of 150 mM of sodium bicarbonate in Ringer's solution. In the animals with remnant stage 3 kidneys, the ratio of absolute bicarbonate reabsorption/absolute sodium reabsorption was increased compared to dogs with stage 1 kidneys at both the proximal and distal sampling sites. These data suggest that bicarbonate reabsorption was elevated in both the distal and the proximal tubules in experimental chronic renal failure. In contrast to the reabsorption of bicarbonate, chloride reabsorption was depressed in stage 3 kidneys at the late proximal puncture site. This resulted in greater delivery to the distal nephron. The distal segments reabsorbed a constant fraction of delivered chloride, resulting in an increase in fractional chloride excretion in chronic renal failure compared to that in normal animals.

Réabsorption tubulaire des bicarbonates chez des chiens en insuffisance rénale expérimentale. Dans cette étude, des expériences de microponction ont été effectuées pour examiner la réabsorption segmentaire de bicarbonates et de chlorures dans le rein de chien normal (stade 1) ou dans le rein restant (stade 3) de chiens en insuffisance rénale expérimentale. Le protocole a comporté trois phases: *(1)* expansion de 3% du liquide extracellulaire, *(2)* perfusion de solution de Ringer contenant 90 mM de bicarbonate de sodium, *(3)* perfusion de 150 mM de bicarbonate de sodium dans de la solution de Ringer. Chez les animaux avec le rein restant au stade 3, le rapport de la réabsorption absolue de bicarbonate/réabsorption absolue de sodium était augmenté par rapport aux chiens avec des reins de stade 1, aux sites de recueil proximaux et distaux. Ces données suggèrent que la réabsorption des bicarbonates était élevée dans les tubules proximaux et distaux lors d'insuffisance rénale chronique expérimentale. A l'opposé de la réabsorption des bicarbonates, la réabsorption des chlorures était diminuée dans les reins au stade 3 aux sites de ponction proximaux tardifs. Cela entrainait une plus grande charge délivrée au néphron distal. Les segments distaux réabsorbaient une fraction constante des chlorures délivrés, ce qui aboutissait à une augmentation de l'excrétion fractionnelle de chlorures lors de l'insuffisance rénale chronique, par rapport aux animaux normaux.

Early studies in uremic humans and rats demonstrated a depression in renal bicarbonate absorption as kidney mass is reduced [1–5]. In contrast, more recent experiments in dogs [6, 7] and rats [8] with experimental renal failure suggest that bicarbonate reabsorption is enhanced. Micropuncture studies in normal rats and dogs have indicated that the early postglomerular segment of the proximal convoluted tubule preferentially reabsorbs sodium bicarbonate relative to sodium chloride [9–13]. Accordingly, the preferential reabsorption of bicarbonate provides the osmotic driving force for salt and water reabsorption and the resulting chloride gradient provides the potential difference which would facilitate sodium movement [14, 15]. Fractional sodium reabsorption falls progressively with reduction in renal mass [16]. It may be speculated that if sodium bicarbonate reabsorption is enhanced under these conditions [6–8], fractional sodium chloride absorption must be diminished. The present micropuncture experiments were designed to study the segmental handling of sodium, bicarbonate, and chloride in dogs with experimental renal failure.

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Methods

Recollection micropuncture experiments were performed on 19 mongrel dogs of both sexes. Eight normal dogs served as stage 1 animals (two normal intact kidneys). Remnant kidneys were induced with segmental infarction in 11 dogs by ligating three-fourths to five-sixths of the main branches of the renal artery of the left kidney. Two weeks after the initial surgery, the right kidney was removed and the animals were studied 1 week later, when they were azotemic. These were designated stage 3 animals. The mean BUN in stages 1 and 3 dogs was 10 ± 1 and 65 ± 10 mg/100 ml, respectively.

All dogs were thyroparathyroidectomized (TPTX) on the day of the micropuncture study to avoid the acute effects of changing PTH levels on renal handling of bicarbonate. Two hours were permitted to elapse following TPTX before standard clearance and micropuncture experiments were performed on the left (normal or remnant) kidney. The dogs were prepared for micropuncture experiments as described previously [17]. Briefly, the animals were anesthetized with intravenous injections of 25 mg/kg sodium pentobarbital, and the level of anesthesia was maintained by injecting small doses of sodium thiopental when necessary. An endotracheal tube was inserted and adequate respiration was maintained with a respirator (Harvard Apparatus Co., Inc., Millis, Massachusetts). The jugular and foreleg veins were cannulated for the infusion of inulin, Ringer's solutions, and sodium bicarbonate. The femoral vein was cannulated to collect blood samples and the femoral artery to

Table 1. Mean blood chemistry data[a]

Stages	Exptl phases	Hct %	P_{Prot} g%	BUN mg%	P_{Cr} mg%	P_{Na} mM	P_{pH}	P_{HCO_3}	P_K	P_{Cl}
								mM		
1	1	35 ± 1	3.88 ± 0.18	9 ± 1	0.86 ± 0.06	153 ± 2	7.32 ± 0.05	17 ± 0.6	3.06 ± 0.12	118 ± 2.3
(N = 8)	2	29 ± 2[b]	2.90 ± 0.23[b]			155 ± 2	7.52 ± 0.05[b,d]	33 ± 0.6[b]	2.35 ± 0.12[b]	109 ± 1.8[b]
	3	28 ± 2	2.61 ± 0.28[b]			156 ± 2[c]	7.71 ± 0.06[b]	43 ± 21.5[b]	2.09 ± 0.07[b]	94 ± 1.7[b]
3	1	36 ± 1	4.61 ± 0.16[d]	65 ± 10[d]	4.49 ± 0.05[d]	153 ± 1	7.23 ± 0.02[d]	18 ± 0.7	3.49 ± 0.16	117 ± 1.4
(N = 11)	2	30 ± 1[b]	3.38 ± 0.17[b]			152 ± 1	7.42 ± 0.02[b,d]	30 ± 0.6[b]	3.15 ± 0.15[b]	106 ± 1.2[b]
	3	28 ± 2[c]	2.90 ± 0.14[b]			153 ± 1	7.55 ± 0.07[b,d]	43 ± 1.0[b]	2.94 ± 0.16[c]	92 ± 1.1[b]

Abbreviations: Hct, hematocrit; BUN, blood urea nitrogen; P, plasma concentration; Cr, creatinine; Prot, protein; HCO_3, bicarbonate; Na, sodium; K, potassium; Cl, chloride.

[a] Values are mean ± SEM.

[b] $P < 0.01$.

[c] $P < 0.05$ compared to previous phase.

[d] $P < 0.01$ compared to corresponding phase in stage 1.

monitor BP. The bladder was exposed by a suprapubic incision, and both ureters were catheterized with polyethylene tubing. The left kidney was exposed through a flank incision and prepared for micropuncture by standard techniques [13].

Recollection micropuncture was used to obtain tubule fluid samples at the same site in all three phases. An oil block of about 40 to 60 μ in length was maintained immediately distal to the puncture site to prevent retrograde collection of tubule fluid. The paraffin oil used in the pipette was equilibrated with a 5% CO_2 gas mixture to achieve a P_{CO_2} of 35 to 45 mm Hg. The last accessible segment of the superficial proximal tubule was chosen for micropuncture to facilitate the comparison of tubular transport between different animal groups. This was done by injection of 4% FD&C green dye (Warren and Jenkinson, St. Louis, Missouri) into the renal artery through a polyethylene cannula (PE 20) and by timing the appearance of the dye in the surface tubules. The distal tubules were identified in a similar manner. Primary and sustaining doses of inulin were administered (1 ml/min) in amounts calculated to provide a stable plasma concentration of about 100 mg/dl. After an equilibration period of 60 min, the first clearance period was initiated. Standard urine collections of 15 min duration were obtained, and blood samples were drawn at the midpoint of each period for determination of sodium, chloride, bicarbonate, and inulin concentrations.

Three phase experiments were performed. Phase 1 consisted of volume expansion with 3% body weight of Ringer's solution (in millimolar concentrations: Na 140, K 3.1, Ca 1.5, Mg 0.5, Cl 128, and HCO_3 20), followed by phase 2 administration of 3% body weight of Ringer's containing 90 mM $NaHCO_3$ (Na 140, K 31, Ca 1.5, Cl 58, Mg 0.5, and HCO_3 90) at an infusion rate of 6 ml/min. Finally, in phase 3 the animals received 3% body weight of Ringer's solution containing 150 mM $NaHCO_3$ and a sustaining infusion at an appropriate rate to replace urinary losses.

Plasma and urine sodium concentrations were determined with flame photometry (Corning 450, Medfield, Massachusetts), and chloride was analyzed by a chloridometer (Buchler-Cotlove, Buchler Instruments, Fort Lee, New Jersey). Plasma and urine inulin was quantitated by the anthrone method of Fuhr, Kaczmarczyk, and Kruttgen [18]. Blood and urine pH and P_{CO_2} were determined at 37°C with the aid of a blood gas analyzer (165/2 pH blood gas analyzer, Corning, Medfield, Massachusetts). The volumes of the tubule fluid collections were determined with the use of constant bore glass capillaries (I.D. 0.33 mm) and a cathetometer (Gaertner Scientific Corp., Chicago, Illinois). Tubule fluid samples were analyzed for inulin by the fluorometric method of Vurek and Pegram [19]. The single nephron glomerular filtration rate (SNGFR) and whole kidney glomerular filtration rate (GFR) were estimated from the single nephron and whole kidney inulin clearances, respectively. Total carbon dioxide concentration was determined by microcalorimetry, according to Vurek, Warnock, and Corsey [20]. Within the pH range of the present experiments, the total carbon dioxide represented bicarbonate plus the dissolved carbon dioxide, carbonate, and carbonic acid. At physiologic pH values, the predominant substance measured was bicarbonate. The sample volume was 13 nl and was introduced into the picapnotherm chamber in most instances immediately following collection. All samples were kept in mineral oil equilibrated to P_{CO_2} of 40 mm Hg and analyzed at this carbon dioxide tension. An electron microprobe (Camebax, Cameca Instruments Inc., Stamford, Connecticut) was used for the analysis of tubule fluid sodium and chloride concentrations using methods described previously [21].

Statistical analysis of paired micropuncture data was carried out by the Student's t test on values obtained during the control and experimental phases in all animals. Standard formulae were used to calculate fractional and absolute reabsorption rates in the various nephron segments [21]. Absolute bicarbonate and chloride reabsorption in the proximal tubule and Henle's loop were determined by the rates delivered to the late superficial proximal tubule and distal collection sites and were based on the mean SNGFR for that animal. To assess significant changes in bicarbonate reabsorption, absolute bicarbonate reabsorption was factored by absolute sodium reabsorption in the proximal and distal tubules at each plasma bicarbonate concentration. The corresponding regressions of reabsorption versus plasma bicarbonate were statistically compared in assessing changes between stages 1 and 3, using covariance analysis.

Results

Clearance data. The mean data for the blood chemistry in stages 1 and 3 dogs are given in Table 1. In stage 3 dogs the BUN and plasma creatinine concentrations were significantly elevated. Plasma hematocrit and protein concentration fell by

Table 2. Mean clearance data from stages 1 and 3[a]

Stage	Exptl phases	V	GFR	FE_{H_2O}	FE_{Na}	FE_K	FE_{HCO_3}	FE_{Cl}
		ml/min				%		
1	1	2.2 ± 0.6	18.0 ± 1.5	12 ± 3	8 ± 2	36 ± 5	4 ± 1	11 ± 2
	2	4.8 ± 0.7[b]	17.5 ± 1.6	27 ± 4	19 ± 3[b]	74 ± 6	35 ± 3[b]	18 ± 3[b]
	3	5.4 ± 0.8	13.6 ± 1.2[b]	39 ± 3	28 ± 3[b]	105 ± 7[b]	62 ± 3[b]	18 ± 3
3	1	1.2 ± 0.3	4.5 ± 0.6[d]	28 ± 4[d]	18 ± 3[d]	95 ± 14[d]	4 ± 1	24 ± 3[d]
	2	1.8 ± 0.3[b,d]	4.6 ± 0.6[d]	42 ± 4[b,d]	30 ± 4[b,c]	123 ± 14[b,d]	32 ± 4[b]	35 ± 4[b,d]
	3	1.4 ± 0.3[b,d]	3.7 ± 0.6[d]	40 ± 4	29 ± 3	154 ± 18[b,d]	47 ± 3[b,d]	26 ± 4[c,e]

Abbreviations: V, urine volume; GFR, glomerular filtration rate; FE_{H_2O}, FE_{Na}, FE_{HCO_3}, FE_{Cl}, fractional excretion of water, sodium, bicarbonate, and chloride, respectively.
[a] Values are mean ± SEM.
[b] $P < 0.01$.
[c] $P < 0.05$ compared to the previous phase.
[d] $P < 0.01$.
[e] $P < 0.05$ compared to the corresponding phase in stage 1.

Table 3. Overall kidney absolute reabsorption for various stages[a]

Stage	Exptl phase	F Na	F HCO_3	F Cl	Abs Na R	Abs HCO_3R	Abs Cl R	Abs HCO_3R	Abs Cl R
		μmoles/min						Abs Na R %	Abs Na R %
1	1	2736 ± 223	305 ± 27	2128 ± 306	2517 ± 222	293 ± 26	1869 ± 149	11 ± 0.6	74 ± 0.8
	2	2740 ± 257	569 ± 51[b]	2078 ± 278[c]	2231 ± 228[b]	374 ± 44[b]	1543 ± 141[b]	16 ± 0.7[b]	69 ± 0.9[b]
	3	2157 ± 209[b]	682 ± 70[b]	1160 ± 191[b]	1548 ± 146[b]	254 ± 32[b]	1545 ± 77[b]	16 ± 0.8[c]	67 ± 1.1[b]
3	1	690 ± 93[d]	81 ± 11[d]	529 ± 74[d]	568 ± 77[d]	78 ± 10[d]	400 ± 56[d]	14 ± 0.7[d]	71 ± 1.2[d]
	2	709 ± 93[d]	140 ± 19[b,d]	482 ± 63[c,d]	513 ± 80[b,d]	99 ± 15[b,d]	325 ± 51[b,d]	19 ± 0.6[b,d]	65 ± 0.9[b,d]
	3	558 ± 98[b,d]	156 ± 11[c,d]	334 ± 56[b,d]	403 ± 73[b,d]	84 ± 15[c,d]	249 ± 49[b,d]	21 ± 0.7[b,d]	61 ± 1.9[b,d]

Abbreviations: F, filtered load; Abs, the absolute reabsorption.
[a] Values are the mean ± SEM.
[b] $P < 0.01$.
[c] $P < 0.05$ compared to the previous phase.
[d] $P < 0.05$ compared to the corresponding phase in stage 1.

Table 4. Mean proximal micropuncture data[a]

Stage	Exptl phases	TF/P_{In}	TF/P_{Na}	$TF/P_{Total\ CO_2}$	TF/P_{Cl}	RF_{Na}	RF_{CO_2}	RF_{Cl}
							%	
1	1	1.67 ± 0.07	1.00 ± 0.01	0.59 ± 0.03	1.10 ± 0.04	60 ± 2	36 ± 2	66 ± 4
	2	1.64 ± 0.08	1.00 ± 0.08	0.83 ± 0.03[b]	1.04 ± 0.03	62 ± 3	51 ± 4[b]	64 ± 3
	3	1.48 ± 0.08[b]	0.99 ± 0.02	0.97 ± 0.02[b]	0.98 ± 0.04	71 ± 3[b]	70 ± 3[b]	72 ± 3
3	1	1.28 ± 0.04[d]	0.99 ± 0.01	0.59 ± 0.03	1.13 ± 0.01	75 ± 2[d]	45 ± 2[d]	84 ± 1[d]
	2	1.24 ± 0.04[d]	0.99 ± 0.01	0.76 ± 0.05[b]	1.07 ± 0.01[b]	78 ± 2[d]	63 ± 3[b,d]	84 ± 1[d]
	3	1.23 ± 0.04[d]	0.98 ± 0.02	0.85 ± 0.04[d]	1.02 ± 0.01[b]	78 ± 2[d]	68 ± 3[c]	83 ± 1[d]

Abbreviations: TF/P In, Na, total CO_2, Cl, tubule fluid to plasma concentration ratio of inulin, sodium, total carbon dioxide, and chloride; RF, fraction remaining.
[a] Values are the mean ± SEM.
[b] $P < 0.01$.
[c] $P < 0.05$ compared to the previous phase.
[d] $P < 0.01$.
[e] $P < 0.05$ compared to the corresponding phase in stage 1.

similar amounts in both stages 1 and 3 animals during bicarbonate infusion. Plasma bicarbonate concentration progressively rose stepwise, accompanied by a decrease in plasma chloride following sodium bicarbonate infusion. The changes in plasma bicarbonate and chloride concentrations in the experimental phases were similar in the two groups of dogs. Plasma sodium concentration remained constant throughout the experiments.

Table 2 summarizes the overall clearance data obtained from these experiments. Stage 3 dogs had a lower GFR than stage 1. Fractional water, sodium, and chloride excretions were significantly higher in stage 3 dogs than in stage 1 animals. There was no difference in fractional bicarbonate excretion between stages 1 and 3 in phases 1 and 2. Table 3 summarizes the absolute reabsorption of sodium, bicarbonate, and chloride. At comparable plasma levels, the absolute bicarbonate reabsorption/absolute sodium reabsorption rate was significantly elevated in stage 3 compared to stage 1, whereas the absolute chloride reabsorption/absolute sodium reabsorption rate was significantly re-

Table 5. Mean distal micropuncture data[a]

Stage	Exptl phases	TF/P_{In}	TF/P_{Na}	$TF/P_{Total\ CO_2}$	TF/P_{Cl}	RF_{Na}	RF_{CO_2}	RF_{Cl}
						%		
1	1	6.11 ± 0.08[b]	0.29 ± 0.02	0.25 ± 0.04	0.36 ± 0.02	5 ± 1	5 ± 1	6 ± 1
	2	3.27 ± 0.44	0.45 ± 0.02[b]	0.73 ± 0.05[b]	0.51 ± 0.03[c]	12 ± 2[b]	27 ± 2[b]	16 ± 2[b]
	3	2.78 ± 0.48[b]	0.47 ± 0.02	0.93 ± 0.13[b]	0.49 ± 0.04	19 ± 2[b]	39 ± 4[b]	22 ± 3
3	1	2.70 ± 0.26[d]	0.33 ± 0.02	0.21 ± 0.05	0.33 ± 0.03	13 ± 1[d]	8 ± 1[d]	13 ± 1[d]
	2	2.36 ± 0.16[d]	0.42 ± 0.02[b]	0.50 ± 0.03[b,d]	0.43 ± 0.03[b,d]	18 ± 1[b,d]	21 ± 1[b,d]	18 ± 1[b]
	3	2.18 ± 0.20	0.46 ± 0.02	0.79 ± 0.05	0.42 ± 0.02	21 ± 1[c]	36 ± 2[b]	19 ± 1

Abbreviations: TF/P_{In}, Na, total CO_2, Cl, tubule fluid to plasma concentration ratio of inulin, sodium, total carbon dioxide and chloride; RF, fraction remaining.

[a] Values are the mean ± SEM.

[b] $P < 0.01$.

[c] $P < 0.05$ compared to the previous phase.

[d] $P < 0.01$.

[e] $P < 0.05$ compared to the corresponding phase in stage 1.

Table 6. Absolute reabsorption of bicarbonate at various nephron segments

Stages	Exptl phases	SNGFR	Filtered load	Abs proximal R	Loop delivery	Abs loop R
			pmoles/min			
1	1	48 ± 4	808 ± 67	511 ± 38	295 ± 34	94 ± 14
	2	51 ± 5	1667 ± 149[a]	816 ± 99[a]	850 ± 81[a]	233 ± 52[a]
	3	44 ± 4[a]	2213 ± 227[a]	687 ± 118	1525 ± 137[a]	477 ± 59[a]
3	1	69 ± 4[c]	1264 ± 120[c]	711 ± 91[d]	577 ± 56[c]	204 ± 16[c]
	2	69 ± 4[c]	2043 ± 154[a,d]	884 ± 122	1172 ± 72[a,c]	525 ± 21[b,c]
	3	68 ± 3[c]	2947 ± 183[a,d]	1085 ± 160[b]	1958 ± 125[a,c]	611 ± 95

Abbreviations: SNGFR, single nephron glomerular filtration rate; Abs, absolute; R, reabsorption.

[a] $P < 0.01$.

[b] $P < 0.05$ compared to the previous phase.

[c] $P < 0.01$.

[d] $P < 0.05$ compared to the corresponding phase in stage 1.

duced in stage 3 compared to stage 1. Accordingly, the reduction in fractional sodium reabsorption in chronic renal failure is related to a decrease in sodium chloride reabsorption rather than a decrease in sodium bicarbonate reabsorption.

Micropuncture data. The micropuncture data obtained from these experiments are summarized in Tables 4 to 6. The mean proximal tubule fluid to plasma concentration ratio (TF/P) of inulin ratio in stage 3 dogs was 1.28 (Table 4), which was significantly lower than the corresponding phase in stage 1 animals, reflecting a reduction in fractional water reabsorption. The mean TF/P sodium ratio remained near unity in all experiments. The proximal TF/P carbon dioxide ratio rose similarly in all stages in response to bicarbonate infusion, whereas TF/P chloride fell following bicarbonate infusion. When the data are expressed as the percentage of filtered load remaining, more sodium, bicarbonate, and chloride remained at the end of the proximal nephron of stage 3 dogs than of stage 1 animals.

Table 5 summarizes the mean distal micropuncture data. Like that of the proximal tubule, the mean distal TF/P inulin ratio was reduced in stage 3 dogs compared to the normal animals. The fraction of filtered sodium bicarbonate and sodium chloride remaining at the distal sampling site was higher in stage 3 than in the corresponding phase in stage 1. Absolute bicarbonate reabsorption rates by the various nephron sites are given in Table 6. It can be seen that absolute bicarbonate reabsorption was higher in stage 3 than in stage 1 in the initial phase, and, following bicarbonate loading, absolute bicarbonate reabsorption was higher in stage 3; this was statistically significant in the loop segment. Figures 1 to 3 illustrate the absolute bicarbonate reabsorption by the various nephron segments. Figure 1 illustrates the absolute bicarbonate reabsorption/absolute sodium reabsorption ratio during bicarbonate loading in the proximal tubule. Bicarbonate reabsorption was increased in stage 3 relative to stage 1 animals. Comparable results were obtained from the distal sampling site (Fig. 2). As shown in Figure 3, at plasma bicarbonate levels greater than about 18 mM, the ratio of absolute bicarbonate reabsorption to absolute sodium reabsorption rate was significantly higher in the azotemic stage 3 dogs. The data suggest that the maximal rate of bicarbonate reabsorption increases in the azotemic dogs and that this increase occurs in both proximal and distal tubules. The loop segment in the present experiments encompasses all the segments between the last accessible proximal tubule and the distal tubule sampling sites. Absolute chloride reabsorption in the proximal tubule is reduced in stage 3 (Table 7), despite the higher filtered load due to an enhanced SNGFR. As a consequence of depressed chloride reabsorption in the proximal nephron, the amount of chloride delivered into the loop segment is markedly increased in stage 3 dogs. Although a large fraction of chloride delivered out of the proximal tubule was reabsorbed by the loop segment, a substantial amount escaped reabsorption and resulted in an increase in fractional excretion of chloride in the stage 3 dogs.

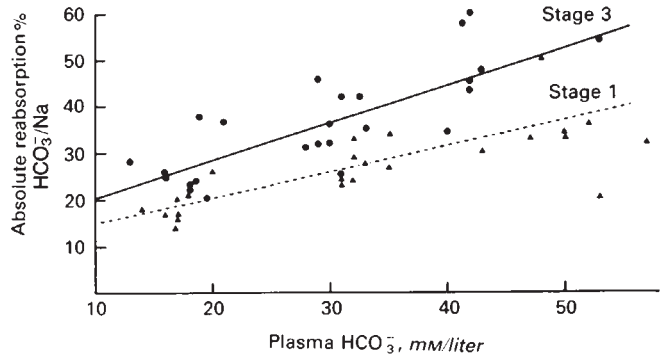

Fig. 1. *Absolute bicarbonate reabsorption and absolute sodium reabsorption ratio in the proximal tubule during graded bicarbonate infusion in both stages 1 (▲, control) and 3 (●, experimental renal failure) dogs. Regression lines* for stage 1 dogs are $y = 0.52 x + 10.26$ ($r = 0.82$, $P < 0.01$) and for stage 3 dogs $y = 0.83 x + 11.68$ ($r = 0.86$, $P < 0.01$). Comparison of the *linear regression lines* indicates significant differences between stages 1 and 3 (slope < 0.01, intercept $P < 0.05$).

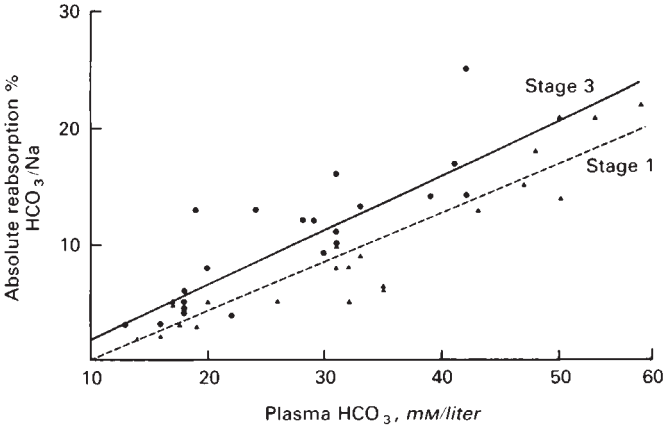

Fig. 2. *Absolute bicarbonate reabsorption in the loop of Henle during graded bicarbonate infusion in stages 1 and 3 dogs. Regression lines* for stage 1 (▲) dogs are $y = 0.45 x - 4.67$ ($r = 0.93$, $P < 0.01$) and for stage 3 (●) dogs $y = 0.50 x - 4.23$ ($r = 0.87$, $P < 0.01$). Comparison of the *linear regression lines* indicates significant differences between stages 1 and 3. The *intercept* is $P > 0.05$ and the *slope* $P < 0.01$.

Discussion

Clearance experiments in rats and dogs have demonstrated avid tubular bicarbonate reabsorption in chronic renal failure [6–8]. In the present micropuncture study, we have examined segmental bicarbonate reabsorption in dogs with experimental renal failure to assess bicarbonate reabsorption along the length of the superficial proximal tubule and loop of Henle. Our clearance and micropuncture results agree with previous studies [6–8], demonstrating that the ratio of absolute bicarbonate reabsorption/absolute sodium reabsorption is increased in chronic renal failure in the superficial proximal (Fig. 1) and, more importantly prior to the distal sampling sites (Fig. 2).

The tubule fluid obtained from the distal sampling site represents fluid which was delivered out of the superficial convoluted tubule and has traversed the straight proximal segments, the thin descending and the thick ascending limb of Henle's loop. Since a major fraction of the bicarbonate delivered into this segment is reabsorbed by the straight proximal tubule [22] and negligible amounts by the ascending limb, the changes in bicarbonate reabsorption detected at the distal sampling site probably reflect alterations in bicarbonate transport in the straight proximal tubule. Figure 2 demonstrates the ratio of bicarbonate over sodium reabsorption obtained at the distal site relative to plasma bicarbonate. This ratio appears to be directly proportional to the plasma bicarbonate concentration in both normal and diseased kidneys, and the ratio is higher in stage 3 than stage 1 dogs. These data suggest that mean bicarbonate reabsorption is elevated in the distal tubule as well as the proximal tubule in chronic renal failure.

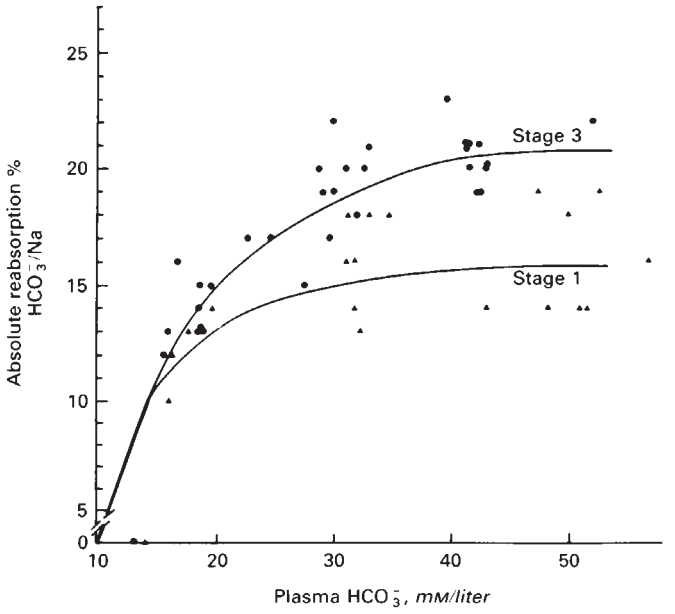

Fig. 3. *Absolute bicarbonate reabsorption by the whole kidney during graded bicarbonate infusion as a fraction of the absolute sodium reabsorption in stages 1 (▲) and 3 (●).*

Several associated factors must be considered in any situation in which bicarbonate reabsorption is augmented. Bicarbonate reabsorption is augmented during potassium depletion and decreased during potassium repletion [23]. Intracellular potassium depletion has been observed in chronic renal failure [24], and hence, the increase in bicarbonate reabsorption seen in the present study may be related to changes in total body potassium. Plasma potassium did not change in the present experiments and was elevated in stage 3 dogs. Potassium depletion would not appear to account for the enhanced bicarbonate reabsorption. Phosphate depletion and elevation of plasma phosphate concentration in chronic renal failure may contribute to hyperabsorption of bicarbonate [25]. Schmidt and Gavellas [7] failed to observe a decrease in bicarbonate reabsorption in dogs with chronic renal failure maintained on a restricted phosphate intake, suggesting that plasma phosphate may not be involved in the present tubular changes. In addition, systemic pH may alter bicarbonate reabsorption. More importantly, blood pH may significantly alter tubular bicarbonate absorption. A decrease in blood pH, as is observed in azotemia, increases bicarbonate reabsorption in the remnant kidney. Schmidt, Bricker, and Gavellas [26] failed to demonstrate a

Table 7. Absolute reabsorption of chloride at various nephron segments

Stages	Exptl phases	SNGFR	Filtered load	Abs proximal R	Loop delivery	Abs loop R
			pmoles/min			
1	1	48 ± 4	5646 ± 445	1820 ± 227	3825 ± 513	2446 ± 514
	2	51 ± 5	5624 ± 581	2042 ± 294	3582 ± 405	1771 ± 292[b]
	3	44 ± 4[a]	4152 ± 345[a]	1219 ± 197[a]	2930 ± 185[a]	1450 ± 86
3	1	69 ± 4[c]	7935 ± 586[c]	1308 ± 184[d]	6722 ± 505[c]	4694 ± 347[c]
	2	69 ± 4[c]	7254 ± 535[d]	1170 ± 144[c]	5988 ± 398[b,c]	4020 ± 321[a,c]
	3	68 ± 3[c]	6365 ± 385[a,c]	1124 ± 139	5234 ± 263[a,c]	3311 ± 119[a,c]

Abbreviations: SNGFR, single nephron glomerular filtration rate; Abs, absolute; R, reabsorption.
[a] $P < 0.01$.
[b] $P < 0.05$ compared to the previous phase.
[c] $P < 0.01$.
[d] $P < 0.05$ compared to the corresponding phase in stage 1.

difference in blood pH between control dogs and animals with experimental renal failure despite the significant increase in bicarbonate reabsorption. Another causative factor may be an increase in the single nephron filtration rate, which is seen in chronic renal failure animals. Recent studies by Corman et al [27] suggest that the postglomerular segment of the proximal tubule, where the bulk of filtered bicarbonate is reabsorbed, may extend further beyond the early proximal tubule in response to an increase in filtered load. Hence, hyperfiltration may contribute to an augmentation in bicarbonate reabsorption by the process of segmental recruitment. At comparable plasma bicarbonate concentrations, there is an adaptive increase in the single nephron filtration rate in the animals, and in the present experiments the absolute bicarbonate reabsorption ratio was higher in stage 3 animals than in stage 1 (Fig. 1). Accordingly, the increase in bicarbonate reabsorption in the azotemic animals may be related to hyperfiltration. Further studies are required to determine the role of these factors in the renal adaptation of bicarbonate absorption in chronic renal failure.

In contrast to bicarbonate, chloride reabsorption is depressed in stage 3 animals at both the proximal and distal puncture sites. In the proximal tubule absolute chloride reabsorption is significantly lower in stage 3 than in stage 1; and similarly, the fraction of sodium remaining is higher in stage 3 animals. Thus, sodium bicarbonate is reabsorbed preferentially to sodium chloride in the proximal tubule of stage 3 dogs. The reduction in sodium chloride reabsorption in the proximal nephron results in an increase in delivery into the loop segments. The fractional reabsorption was 60 ± 4 and 70 ± 2% in stages 1 and 3, respectively. It would appear that a constant fraction of chloride delivered out of the proximal tubule is reabsorbed by the loop segments independent of loop delivery. The increase in overall fractional chloride excretion in stage 3 is due to a reduction in chloride reabsorption in the proximal tubular segments, which are not fully compensated for by augmented loop reabsorption.

In summary, the results are compatible with overall clearance experiments indicating an enhancement of tubular bicarbonate reabsorption. The present micropuncture study indicates that bicarbonate reabsorption by the proximal and the distal nephron is increased during chronic reduction in renal mass. The augmentation in chloride excretion in renal failure is presumably due to an increase in proximal bicarbonate absorption with an appropriate reduction in chloride reabsorption. Adaptation to reduced nephron mass appears to involve enhanced sodium bicarbonate reabsorption with reduced sodium chloride transport.

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